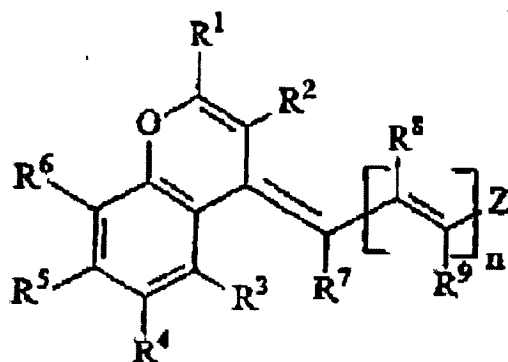


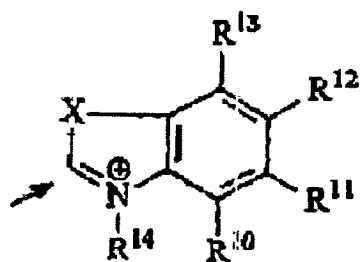
# PATENT CLAIMS

1. Marker dyes based on unsymmetrical polymethines which contain a substituted  $\omega$ -(benz[b]pyran-4-ylidene)alk-1-enyl unit of general structure (I),

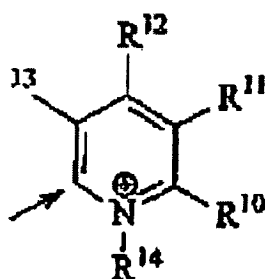


I

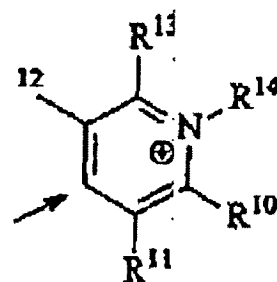
where Z takes the place of substituted derivatives of benzooxazolium, benzothiazolium, 3,3-dimethyl-4,5benzo-3H-indolium, 2- and 4-pyridinium, 2- and 4-chinolinium and 9-acridinium with the general formulae IIa or IIb or IIc



IIa



IIb



IIc

and where

- X stands for an element of the group O, S, Se or the structural element N-alkyl or C(alkyl)<sub>2</sub>,

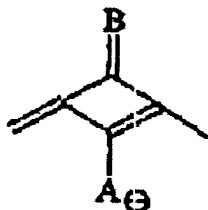
- n stands for the numerical value 0, 1, 2 or 3,
- $R^1$ - $R^{14}$  are equal or different and can be hydrogen, one or more alkyl, or aryl, heteroaryl or heterocycloaliphatic fragments, a hydroxy or alkoxy group, an alkyl-substituted or cyclical amine function and/or two fragments in ortho position to each other, for example  $R^{10}$  and  $R^{11}$ , can together form another aromatic ring,
- At least one of the substituents  $R^1$ - $R^{14}$  can be a solubilizing or ionizable or ionized substituent, like cyclodextrine, sugar,  $SO_3^-$ ,  $PO_3^{2-}$ ,  $COO^-$ , or  $NR_3^+$ , which determines the hydrophilic properties of these dyes; here it is possible that this substituent can be bound to the marker dye by means of a spacer group,
- At least one of the substituents  $R^1$ - $R^{14}$  can stand for a reactive group which facilitates a covalent linking of the dye another carrier molecule, while this substituent can also be bound to the dye by means of a spacer group, and
- $R^1$  is a substituent which has a quarternary C-atom in alpha-position relative to the pyran ring, wherein the substituents  $R^1$  and  $R^2$  can also form an aliphatic or a substituted aliphatic ring system.

2. Laser-compatible NIR marker dyes according to claim 1, characterized in that the reactive group has been chosen from one of the following functionalities: Isothiocyanates, isocyanates, monochlorotriazines, dichlorotriazines, aziridenes, sulfonylhalogenides, carbonic acid chlorides, *N*-hydroxysuccinimideesters, imido-esters, glyoxal or aldehyde for amine and hydroxy functions or maleimides or iodoacetamides for thiol functions and phosphoramidites for the marking DNA or RNA or their fragments.

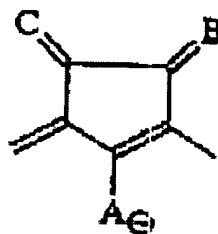
3. Laser-compatible NIR marker dyes according to claims 1-2, characterized in that the reactive group is bound to the actual chromophore by means of spacer groups of the general structure  $-(CH_2)_m$ , where m can have a value from 1 to 18.

4. Laser-compatible NIR marker dyes according to claims 1-3, characterized in that the structural unit =CR8- also contains a bridging via four-, five- and six-membered ring systems; on it there are also reactive groups and the substituents A – G have the same functionality as the substituents  $R^1 - R^{14}$ .

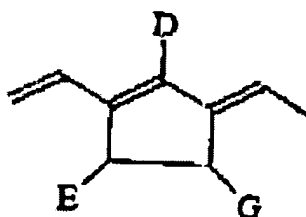
5. Laser-compatible NIR marker dyes according to claim 4, characterized in that the structural unit  $=CR^8-$  stands for



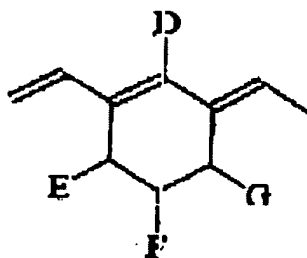
6. Laser-compatible NIR marker dyes according to claim 4, characterized in that the structural unit  $=CR^8-$  stands for



7. Laser-compatible NIR marker dyes according to claim 4, characterized in that the structural unit  $=CR^8-$  stands for



8. Laser-compatible NIR marker dyes according to claim 4, characterized in that the structural unit =CR8- stands for



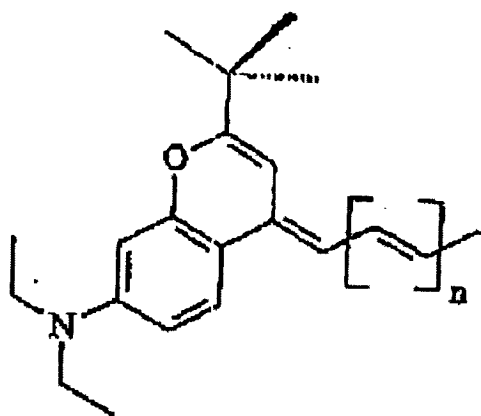
9. Laser-compatible NIR marker dyes according to claims 1-8, characterized in that the substituents A – G can be identical to the substituents  $R^1$  to  $R^{14}$ , or stand for O, S,  $C(CN)_2$  or N-R, where the R in N-R can stand for an aliphatic or aromatic or a reactive aliphatic or aromatic fragment, like  $(CH_2)_nCOOH$  or  $(CH_2)_nNH_2$ .

10. Laser-compatible NIR marker dyes according to claims 1-8, characterized in that the substituent D to which possibly reactive substituents corresponding to the substituents  $R^1$  to  $R^{14}$  are attached stands for Cl, an alkyl, alkoxy, cycloalkoxy, phenolate, alkylmercapto or phenylmercapto fragment.

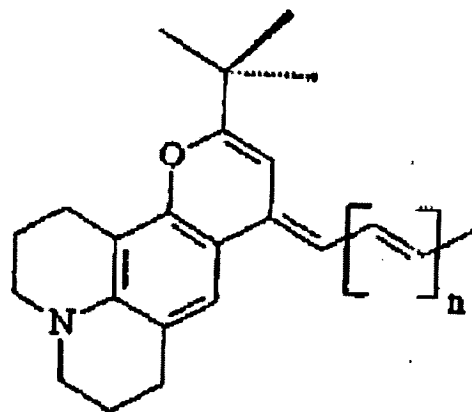
11. Laser-compatible NIR marker dyes according to claim 1, characterized in that the substituent  $R^1$  stands for 1,1-dimethylethyl, 6,6-dimethylbicyclo-[3,1,1]-hept-2-en-2-yl, bicyclo[2,2,1]-hept-2-en-5-yl, 3,3-dimethylbut-1-en-1-yl or adamant-1-yl ( $-C_{10}H_{15}$  / tricyclo[3.3.1.1<sup>3,7</sup>]decyl).

12. Laser-compatible NIR marker dyes according to claim 1, characterized in that the substituent  $R^5$  stands for a singly or doubly alkylated amine, wherein alkyl and alkenyl bridgings with the substituents  $R^4$  and  $R^6$  are possible via the aminonitrogen.

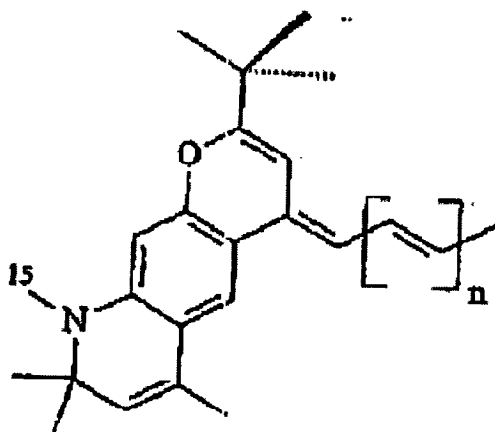
13. Laser-compatible NIR marker dyes according to claims 1-12, characterized in that the  $\omega$ -(benz[b]pyran-4-ylidene)alk-1-enyl) part of the dyes has the following structure Ia, Ib, Ic or Id



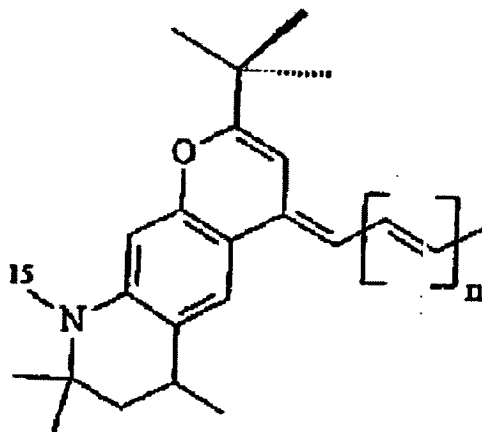
Ia



Ib



Ic



Id

where the substituent  $R^{15}$  can have the same functionalities as the substituents  $R^1$  to  $R^{14}$ .

14. Laser-compatible NIR marker dyes according to claim 1-13, characterized in that

a.  $R^2, R^3, R^4, R^6, R^7, R^8, R^9$  are equal to H,  $R^1$  is equal to 1,1-dimethylethyl,  $R^5$  is equal to N,N-diethylamino,  $n = 1$ , Z is equal to IIa with  $R^{10}, R^{11}, R^{13}$  equal to H,  $R^{12}$  is equal to  $SO_3^-$  and  $R^{14}$  is equal to 5-carboxypent-1-yl, that is 1-(5-carboxypent-1-yl)-3,3-dimethyl-2-[3-(7-N,N-diethylamino-2-(1,1-dimethylethyl)-4H-benzopyran-4-ylidene)-1-propen-1-yl]-3H-indolium-5-sulfonate (OB11),

b.  $R^2, R^3, R^7, R^8, R^9$  are equal to H,  $R^4, R^5, R^6$  together form a julolidine ring,  $R^1$  is equal to 1,1-dimethylethyl,  $n = 1$ , Z is equal to IIa with  $R^{10}, R^{11}, R^{13}$  are equal to H,  $R^{12}$  is equal to  $SO_3^-$  and  $R^{14}$  is equal to 5-carboxypent-1-yl, that is 1-(5-carboxypentyl)-3,3-dimethyl-2-[3-(11-(2,2-dimethylethyl)1H,2H,3H,5H,6H,7H-pyrano[2,3-f]pyrido [3,2,1-ij]chinolin-9-ylidene)-1-propenyl]-3H-indolium-5-sulfonate (OB 15),

c.  $R^2, R^3, R^4, R^6, R^7, R^8, R^9$  are equal to H,  $R^1$  is equal to 1,1-dimethylethyl,  $R^5$  is equal to N,N-diethylamino,  $n = 1$ , Z is equal to IIc with  $R^{10}, R^{11}$  equal to H,  $R^{12}$  and  $R^{13}$  together form a ring which is attached by a condensation reaction and has the structure  $CH=C(SO_3^-)-CH=CH$ , and  $R^{14}$  is equal to 5-carboxypent-1-yl, that is 1-(5-carboxypentyl)-4-[3-(7-N,N-diethylamino-2-(1,1-dimethylethyl)-4H-benzopyran-4-ylidene)-1-propen-1-yl]-chinolinium-6-sulfonate (OB 20),

d.  $R^2, R^3, R^4, R^6, R^7, R^8, R^9$  are equal to H,  $R^1$  is equal to 1,1-dimethylethyl,  $R^5$  is equal to N,N-diethylamino,  $n = 1$ , Z is equal to IIc with  $R^{10}, R^{11}$  equal to H,  $R^{12}$  and  $R^{13}$  together form a ring which is attached by a condensation reaction and has the structure  $CH=CH-CH=CH$ , and  $R^{14}$  is equal to 3-hydroxyprop-1-yl, that is (1-(3-hydroxyprop-1-yl)-4-[3-(7-N,N-diethylamino-2-(1,1-dimethylethyl)-4H-benzopyran-4-ylidene)-1-propen-1-yl]-chinolinium-perchlorate (OB 14)

15. Use of the substituted pyran derivatives with the general formula I as dyes for the optical marking of proteins, nucleic acids, oligomers, DNA, RNA, biological cells, lipids, polymers, drugs or polymer particles.

16. System for the qualitative or quantitative determination of proteins, nucleic acids, oligomers, DNA, RNA, biological cells, lipids, polymers, drugs or polymer particles, characterized in that the functional groups of the compounds according to claims 1

to 14 are coupled covalently to an OH, NH<sub>2</sub> or SH function of the substances to be determined.

17. System according to claim 16, characterized in that the coupling reaction takes place in an aqueous solution.

18. System according to claim 16 or 17, characterized in that the covalently coupled compound has fluorescent properties.

19. Use of the compounds and systems according to claims 1 to 18 in optical, in particular, fluorescence optical qualitative and quantitative determination methods, including immune tests, hybridization methods, chromatographic or electrophoretic methods and high-throughput screenings.

20. Use of the compounds and systems listed in claims 1 to 18 in the analysis of the interaction between receptors and ligands on a microarray.